Review

Epidural electrical stimulation for spinal cord injury

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Abstract

A long-standing goal of spinal cord injury research is to develop effective repair strategies, which can restore motor and sensory functions to near-normal levels. Recent advances in clinical management of spinal cord injury have significantly improved the prognosis, survival rate and quality of life in patients with spinal cord injury. In addition, a significant progress in basic science research has unraveled the underlying cellular and molecular events of spinal cord injury. Such efforts enabled the development of pharmacologic agents, biomaterials and stem-cell based therapy. Despite these efforts, there is still no standard care to regenerate axons or restore function of silent axons in the injured spinal cord. These challenges led to an increased focus on another therapeutic approach, namely neuromodulation. In multiple animal models of spinal cord injury, epidural electrical stimulation of the spinal cord has demonstrated a recovery of motor function. Emerging evidence regarding the efficacy of epidural electrical stimulation has further expanded the potential of epidural electrical stimulation for treating patients with spinal cord injury. However, most clinical studies were conducted on a very small number of patients with a wide range of spinal cord injury. Thus, subsequent studies are essential to evaluate the therapeutic potential of epidural electrical stimulation for spinal cord injury and to optimize stimulation parameters. Here, we discuss cellular and molecular events that continue to damage the injured spinal cord and impede neurological recovery following spinal cord injury. We also discuss and summarize the animal and human studies that evaluated epidural electrical stimulation in spinal cord injury.

Key Words: central nervous system; chondroitin sulfate proteoglycans; epidural electrical stimulation; glial scar; gliosis; neural activity; neuromodulation; oligodendrocyte; spinal cord injury

Introduction

Spinal cord injury (SCI) is a devastating event with unforeseen physical and emotional consequences for patients. Up to 90% of SCI cases are caused by trauma including vehicle crashes, sports injuries, falls or violence (Organization, 2013). Non-traumatic SCI cases from vascular, neoplastic or infectious origins are also recently increasing (Organization, 2013). The age distribution of SCI cases is bimodal, with a first peak comprising young adults between 15 and 29 years old and a second peak comprising adults over the age of 64 years (van den Berg et al., 2010).

Depending on the severity and location of the damage to the spinal cord, SCI can be divided into complete or incomplete SCI. A complete SCI causes permanent damage to the injured area, leading to a total lack of sensory and motor functions below the injury level, whereas an incomplete SCI refers to partial damage to the spinal cord. Sensory and motor functions below the injury level are partially preserved in an incomplete SCI. Usually, sensory function is preserved to a greater extent than motor function because the sensory tracts are peripherally located in the spinal cord. If the injured level is high in the cervical spinal cord, the patient would suffer from respiratory complications throughout life. In addition, bowel or bladder dysfunctions could be another concern among patients with SCI. Besides the medical consequences of SCI, an economic burden on the patients is also immense because SCI requires lifelong care (Munce et al., 2016; Backx et al., 2018). The lifetime direct costs of SCI range from $2.1 million to $5.4 million per patient. The recognition of the personal and socioeconomic impacts of SCI has fostered extensive basic and clinical research in SCI.

Over the last three decades, great strides have been made in understanding the pathophysiology of SCI and improving the treatment. Especially, post-trauma management and rehabilitation have improved the prognosis of SCI (Katoh et al., 2019). However, these approaches are still limited to minimizing complications and maximizing residual function rather than restoring impaired motor functions back to normal (Ramer et al., 2014). Pharmacologic agents, transplantation of stem cells and implantation of biomaterials have shown positive outcomes in animal models but translational efforts have yet to achieve promising clinical outcomes (Bydon et

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al., 2014; Lang et al., 2015; Li et al., 2018; Kourgiantaki et al., 2020; Liu et al., 2020; Zhao et al., 2020).

Recovery of motor functions is often limited although it is rated as the highest priority by patients with paraplegia or tetraplegia (Anderson, 2004). To overcome these challenges, researchers have developed numerous strategies to regenerate and reorganize the injured spinal cord. These strategies include bodyweight support system, lower limb exoskeletons, functional electrical stimulation of muscles and neuromodulation of the central nervous system (Borton et al., 2013). Importantly, recent clinical investigations have demonstrated that epidural electrical stimulation (EES) of the spinal cord can restore some volitional movement below the level of SCI (Harkema et al., 2011; Angeli et al., 2014; Angeli et al., 2018). These findings have been further supported by the restoration of independent stepping from complete SCI with EES and task-specific training (Gill et al., 2018; Wagner et al., 2018). Despite a limited number of studies, the positive outcomes of EES on autonomic, cardiovascular, respiratory and motor functions further support that EES holds a potential as a therapeutic intervention after SCI. In this review, we summarize cellular and molecular events associated with SCI and discuss studies assessing EES efficacy in animal models and humans with SCI (**Table 1**).

Search Strategy and Selection Criteria

We used the PubMed and Google Scholar to search the literature published from January 1990 to September 2020 with the search terms including spinal cord injury, epidural electrical stimulation and neuromodulation.

Cellular and Molecular Events Associated with Spinal Cord Injury

The pathophysiology of SCI can be categorized into a primary and secondary phase (Tran et al., 2018; Alizadeh et al., 2019). The primary phase occurs with the initial mechanical insult on the spinal cord, disrupting the blood-spinal cord barrier (BSCB) and rupturing the surrounding blood vessels by an exerted force. In addition, this physical insult directly leads to necrosis or apoptosis of astrocytes, oligodendrocytes and neurons within the epicenter of a lesion (Grossman et al., 2000, 2001). The secondary phase represents the subsequent cellular and biochemical processes, which worsen the injury and hinder regeneration of the damaged axons (Tator and Fehlings, 1991; Rowland et al., 2008).

Primary phase of SCI

The primary phase of SCI often results in dysfunction of BSCB, which provides a microenvironment for the spinal cord parenchyma (Bartanusz et al., 2011). Upon injury, the tight junctions between endothelial cells of BSCB can disintegrate, leading to increased vascular permeability and edema (Mautes et al., 2000). The edema results in uncontrolled release of neurotransmitters, alteration of the water content and imbalance of Mg^{2+} and Na^{+} in the cytoplasm. These homeostatic imbalances eventually cause neuronal oxidative stress, protein aggregation and lipid peroxidation (Garcia et al., 2016). Although the BSCB repairs with time, the formation of tight junctions can still fail because of the decreased expression level of claudins and occludins (Chodobski et al., 2011). Moreover, the release of interleukin (IL)-16 and activation of matrix metalloprotease 9 contributes to persistent permeability of the BSCB (Noble et al., 2002; Mueller et al., 2006; Lee et al., 2016). Later, this leaky BSCB can allow the infiltration of inflammatory cells to the injury site.

Major complications of blood vessel rupture following SCI are intraparenchymal hemorrhage and subsequent ischemia. The gray matter is more susceptible to ischemic damage than the white matter because it has a higher density of capillary beds and contains neurons with high metabolic demand (Tator and Koyanagi, 1997). Upon reperfusion of ischemic lesion, free radicals are rapidly generated through the Fenton reaction, causing membrane damage to neurons and glial cells (Shichiri, 2014). Overall, the extent of the primary phase determines secondary phase severity and clinical outcomes. Thus, it is critical to provide expeditious relief of mechanical compression on the spinal cord and attenuate secondary injury cascades with early surgical decompression (Fehlings et al., 2012; Furlan et al., 2016; Wilson et al., 2017; Badhiwala et al., 2018).

Secondary phase of SCI

The multifaceted pathological process continues for weeks or months following the initial primary injury, causing progressive damage at the site of lesion and spinal cord. The term secondary injury refers to a series of cellular, molecular and biochemical events that cause further damage to the spinal cord and hinder regeneration of the damaged axons. The major events in the secondary phase of SCI include neuroinflammation, formation of cystic cavity and maturation of glial scars (Tran et al., 2018; Alizadeh et al., 2019).

When BSCB is compromised during the primary phase, inflammatory cells can rapidly infiltrate into the spinal cord, which should remain as an immune-privileged site. These cells trigger a release of cytokines such as tumor necrosis factor-alpha, IL-1α, IL-1β and IL-6. The levels of these cytokines reach their peak 6 to 12 hours after the injury and remain up to 4 days following SCI (Nakamura et al., 2003; Kumamaru et al., 2012; Nguyen et al., 2012). The first peripheral immune cells infiltrating into the spinal cord following SCI are neutrophils (Taoka et al., 1997). At the lesion site, neutrophils trigger an inflammatory cascade by activating other immune cells and glia through the release of cytokines and chemokines (Taoka et al., 1997). $CD4^{+}$ cells are also activated following the inflammatory cascade and release cytokines that stimulate B cells (Ankeny et al., 2006). These B cells undergo clonal expansion and release autoantibodies against the injured spinal cord, which causes self-destruction (Ankeny et al., 2006). This finding was further supported by high levels of central nervous system reactive IgM and IgG from serological assessment of patients with SCI (Hayes et al., 2002).

The inflammatory cells can also cause damage to the spinal cord through direct contact rather than through secreting the inflammatory factors. For example, activated macrophages directly interact with dystrophic axons and induce retrograde axonal dieback (Horn et al., 2008; Busch et al., 2009; Kigerl et al., 2009). This finding was supported by the time-lapse microscopy studies demonstrating the retraction of dystrophic growth cones after a direct contact with macrophages (Busch et al., 2009, 2011). Upon injury, microglia constantly patrolling the central nervous system change their morphology by extending their processes to the lesion site (Davalos et al., 2005). The activated microglia play a role in removing cellular debris to seal the lesion, but they also contact the damaged axons and phagocytose the membranes (Wu et al., 2005; Greenhalgh and David, 2014). Collectively, a growing body of evidence has demonstrated the critical role of inflammatory cascades in the pathophysiology of SCI.

The substantial loss of volume due to the progressive cell death in the lesion site gets replaced by a cystic cavity, referred to as syringomyelia (Seki and Fehlings, 2008). The cystic cavity contains extracellular fluid, macrophages and bands of connective tissue. Astrocytes surrounding the cavity proliferate and tightly interweave their processes to wall off the cavity. This structure is referred to as the

Table 1 | **Summary of included studies on human epidural electrical stimulation**

Study	Subject numbers (M/F)	ASIA grade	Complete/ incomplete motor	Injury level range across subjects	Stimulation location	Stimulation parameters	Indication/outcome	Subject numbers with meaningful motor recovery
Angeli et al. (2018)	4 (3/1)	ASIA A:2 ASIA B:2	Complete	C4–T4	L1–S1, L1–S2	2 Hz	Intentional over-ground walking ability	1/4
Gill ML et al. (2018)	1 (1/0)	ASIA A	Complete	T6	T11–L1	15–40 Hz. 210 μs width	Independent standing, bilateral stepping on a treadmill, and assisted stepping over ground	1/1
Grahn et al. (2017)	1 (1/0)	ASIA A	Complete	T6	T11–L1, Lumbosacral	25 and 40 Hz (volitional control/stepping) 15 Hz (standing) 0–6 V, 210 ms width	Stand, step like movement with side lying or BWSS	1/1
Rejc et al. (2017)	4 (4/0)	ASIA A:2 ASIA B:2	Complete	C7–T4	T11–L1	25–60 Hz, 1–9 V	Interference of stand/ step training on progression of motor function	4/4
Rejc et al. (2017)	1 (1/0)	ASIA B	Complete	C7	T11–L1	25–60 Hz, 1–9 V	Standing, stepping, and volitional leg movement after task-EES training	1/1
Lu et al. (2016)	2 (NA)	ASIA B	Complete	C5–6	C4–T1	0.1–10.0 mA, 2–40 Hz, 210 μs width	Hand (grip strength, motor control, and action latency) and arm motor function improvements	2/2
Danner et al. (2015)	10 (7/3)	ASIA A:6 ASIA B:4	Complete	C6–T9	T11–L1	5, 10, 16, 21, 25, 31 and 40 Hz, 1–10 V, 210 ms width	Mapping outputs: muscle co-activations, mixed-synergies, and locomotor-like configurations	NA
Hofstoetter et al. (2015)	8 (6/2)	NA	Complete	C5–T6	T11–L1	5–26 Hz	Modulation of lower-limb output EMG patterns	NA
Rejc et al. (2015)	4 (4/0)	ASIA A:2 ASIA B:2	Complete	C7–T4	T11–L1	25–60 Hz, 1–9 V	Full weight bearing without assistance	4/4
Angeli et al. (2014)	4 (4/0)	ASIA A:2 ASIA B:2	Complete	C6–T6	T11–T12	25 or 30 Hz, sub-movement threshold to above optimal	Voluntary limb, the ankle and toe muscles movements	4/4
Minassian et al. (2013)	7 (NA)	NA	Complete	C4–T10	T10–L1	2–42 Hz, 10 s segments	EMG rhythm differs when stimulation is delivered with step-related sensory feedback	NA
Harkema et al. (2011)	1 (1/0)	ASIA B	Complete	C7–T1	T11–L1	0.5 to 10.0 V and 5 to 40 Hz, respectively, using either a 210 or 450 μs width.	Voluntary movement, standing, and assisted stepping	1/1
Carhart et al. (2004)	1 (1/0)	ASIA C	Incomplete	C5–6	T10–T12	Continuous, charge-balanced, monophasic pulse trains at a frequency of 40–60 Hz, a pulse duration of 800 micros, an amplitude eliciting 50% of sensory and motor thresholds	Improved treadmill and over-ground ambulation	1/1
Minassian et al. (2004)	10 (7/3)	ASIA A:8 ASIA B:2	Complete	C4–T10	T10–L1	2.2, 5–15, and 25–50 Hz	Stepping like movement	NA
Herman et al. (2002)	1 (1/0)	ASIA C	Incomplete	C5–6	T11–L2	20–60 Hz, 0.8 ms width, paresthesia/vibration inducing amplitude	Functional walking	1/1
Dimitrijevic et al. (1998)	6 (3/3)	ASIA A	Complete	C5–T8	L2	25 Hz, 5–9 V, 0.2–0.5 ms width	Stepping like movement	NA

ASIA: American Spinal Injury Association; BWSS: body-weight support system; F: female; M: male; NA: not available; SCI: spinal cord injury.

glial scar, which impedes axonal regeneration and causes a poor functional outcome (Hara et al., 2017). The glial scar often spread rostral and caudal to the lesion. Moreover, astrocytes secrete chondroitin sulfate proteoglycans (CSPGs) that form a biochemical barrier to neurite outgrowth and axonal growth (McKeon et al., 1991). The inhibitory effects of CSPGs on axon growth are mediated through receptor protein tyrosine phosphatase sigma (RPTPσ) and leukocyte common antigen-related phosphatase (Shen et al., 2009). It has been demonstrated that blockade of RPTPσ intracellular peptide promotes axonal regeneration (Lang et al., 2015). Like neurons, oligodendrocyte progenitors are also affected by CSPGs of the glial scar. Although oligodendrocyte progenitors migrate toward the lesion for remyelination, CSPGs of the glial scar can potently inhibit myelination (Andrews et al., 2012; Keough et al., 2016). Thus, it is conceivable that CSPGs contribute to chronic remyelination failure following SCI. Inflammation is one of the most common double-edged swords, requiring a fine balance between initiation and termination. Similarly, while the glial scar prevents the spread of inflammatory processes, it also contributes to the failure of axonal regrowth at the lesion site.

Assessment of Epidural Electrical Stimulation in Spinal Cord Injury Animal Models

Experimental models of SCI

Animal models of SCI have been an invaluable tool for investigating new therapeutic modalities and understanding the pathophysiology of SCI. Historically, animal models of SCI include cats, dogs and monkeys prior to the use of rodents (Kjell and Olson, 2016). Rodents have gained popularity in SCI studies due to the ease of genetic manipulation, rapid development and cost-effectiveness compared to larger non-primates and primates (Kwon et al., 2002; Kjell and Olson, 2016). Larger animals provide the opportunity to evaluate and refine promising therapies that have shown efficacy in rodents. According to a recent systematic review of 2209 studies, the most common species involved in animal models of SCI was the rat (72.4%) (Sharif-Alhoseini et al., 2017). The most common spinal region investigated was thoracic (81%) followed by cervical (12%), lumbar (5.1%), sacral (0.7%) and other (0.7%) (Sharif-Alhoseini et al., 2017). Contusion was the most common approach to induce an injury (41%) followed by transection (32.5%) and compression (19.4%) (Sharif-Alhoseini et al., 2017). A contusion or compression is more pertinent for investigating of pathophysiological changes following SCI since they are the inciting events in most human SCI cases. On the other hand, transection is useful to study the effects of scaffolds, biomaterial and neuromodulation. For this reason, transection was the most common method to induce SCI in animal models for the investigation of neuromodulation (Sharif-Alhoseini et al., 2017).

Animal models of SCI for the assessment of EES have been primarily developed in rats and cats. Modeling of SCIs in the absence of supraspinal influence had been performed by precollicular-postmammillary brainstem transection while recent modeling of SCIs has been achieved mainly through spinal cord transection of the mid to lower thoracic levels (T7–13) in both cats and rats (Iwahara et al., 1992). Another animal model using two separate transections has been developed, wherein a transection at L2, rostral L3, caudal L3, or L4 secondary to a T13 transection helped determine spinal cord regions responsible for spinal locomotion (Langlet et al., 2005).

Effect of EES in SCI animal models

There are multiple sites within the brain and spinal cord that can be stimulated to induce locomotion in a variety of invertebrate and vertebrate species. In 1992, it was first demonstrated that EES of the L1 and L4 levels can induce hindlimb locomotion in decerebrated cats (Iwahara et al., 1992). Such finding was also observed with stimulation at the L4 and L5 levels in decerebrated and spinalized cats (Gerasimenko et al., 2002; Musienko et al., 2007). Studies of epidural stimulation in spinalized or decerebrated rats demonstrated that stimulation of the L2 or S1 level was effective in producing bilateral hindlimb locomotion or locomotor-like activity (Ichiyama et al., 2005; Lavrov et al., 2006, 2008a, b). The most effective frequencies of stimulation in rats had ranged from 30–60 Hz while the optimal frequency in cats ranged from 20–35 Hz (Gerasimenko et al., 2002, 2003; Ichiyama et al., 2005; Lavrov et al., 2008a). The variability between frequencies for effective stimulation seemingly stems from the different optimal frequencies that are needed to activate the necessary spinal circuits to induce locomotor activity (Lavrov et al., 2008a). In cat models that utilized two separate transection sites, with the second site of transection occurring around the mid to lower lumbar segments, stimulation above the site of transection was ineffective in eliciting an effective locomotor response (Gerasimenko et al., 2002, 2003).

The electromyography (EMG) activity recorded in the hindlimbs of rats in response to epidural stimulation had three different responses with an early response, a middle response and a late response (LR). The early response arose as a result of direct stimulation of motor neurons, middle response arose as a result of a monosynaptic response, and the LR as a result of a polysynaptic response (Lavrov et al., 2006). Following spinal cord transection, all three of the responses had gradually increased, with the LR having been initially abolished, but had later reappeared at around the three-week mark. This reappearance coincided with the ability to begin stepping in response to stimulation, pointing to its relation with stepping restoration (Lavrov et al., 2006). Another observation from the EMG activity was the difference in the amount of long latency spikes with there being five to seven spikes while bipedal stepping *versus* a single spike while only standing in a bipedal position. The greater amounts of spikes while stepping appears to be evidence of spinal circuits being activated that contribute to the locomotor performance (Lavrov et al., 2008a).

The role of EES in the reappearance of the LR in EMG activity following transection points to the plasticity of the spinal cord as the reappearance of the LR reflects polysynaptic network restoration. This network restoration appears to be responsible for the reflexes of flexor and cross extension as its reappearance coincides with the regained ability to begin bilateral locomotor activity (Lavrov et al., 2006). As a result of spinal cord shock from transection, the interneuron network responsible for generating stepping movement is inactive. The application of EES is seemingly able to reactivate this network allowing the regeneration of stepping movements (Musienko et al., 2007).

In both cat and rat models, bodyweight support and hindlimb contact with a moving treadmill belt were necessary to produce rhythmic and effective bilateral locomotor activity. The absence of these two adjuncts resulted in an unstable rhythm of activity that was not indicative of effective bilateral locomotion as evidenced by poor stepping when the hindlimbs were not in contact with the moving treadmill and the bilateral EMG activity (Ichiyama et al., 2005); this demonstrates the importance of peripheral feedback. Also, the feedback from afferent dorsal roots and the ascending branches in the dorsal columns plays an essential role in conjunction with EES in both initiating and maintaining the excitability of the stepping generator components (Gerasimenko et al., 2002, 2003). Altogether, although further studies elucidating the mechanism of EES in SCI are needed, preclinical investigations of EES in SCI animal models have provided insight into the efficacy of EES.

Assessment of Epidural Electrical Stimulation in Patients with Spinal Cord Injury

Early indications for EES in humans

Epidural electrical stimulation involves the delivery of current to the dorsal aspect of the spinal cord through surgically implanted electrodes. While electrical therapeutics date back to ancient Rome, the EES surgical procedure was inspired by Wall's and Sweet's initiation of electrical stimulation of relevant peripheral nerves for pain abolition in 1967. However, to minimize chronic pain sensation across more diffuse areas, stimulation was then applied to the dorsal columns of the spinal cord (Shealy et al., 1967). Subsequently, indications expanded to include spasmodic torticollis (Gildenberg, 1978), pain and motor function in patients with multiple sclerosis (Cook, 1976), small artery disease (Dooley and Kasprak, 1976), trunk and limb pain, failed back surgery syndrome, refractory angina pectoris, cardiac X syndrome, limb ischemia, regional pain syndrome, and diabetic nephropathy (Mekhail et al., 2018).

Evidence for EES-induced stepping-like movements absent supraspinal influence

The implications of EES for gait after SCI was inspired by several reports of inducing stepping-like movement with stimulation despite little to no supraspinal influence (Bussel et al., 1996; Dimitrijevic et al., 1998; Minassian et al., 2004). These studies supported the notion of the existence of a central pattern generator in humans, previously identified in animal work. Through stimulation of flexor reflex afferents, it was shown that the spinal neural network in paraplegic patients resembled that of the acute spinal cat dopaminergic network and therefore related to the central pattern generator network (Bussel et al., 1996). The same report summarized the observation of rhythmic contractions of lower limb and trunk extensors in one patient. In the same patient, observed were alternating flexion-extension motor activity modulated by flexor reflex afferents stimulation and rhythmic bursts of extensor motor neurons, which is suggested by prior animal work to be generated by the spinal stepping generator (Bussel et al., 1996). It was subsequently reported that electrical stimulation (25–60 Hz, 5–9 V) of the posterior aspect of the lumbar spine (L2 spinal segment) could lead to stepping-like (stance-and-swing phases) movements of the lower limb and step-like EMG activity in six SCI patients lying supine (Dimitrijevic et al., 1998).

Subsequent characterization of spinal input-output relationships

This early work was followed by a characterization of lower limb muscle EMG correlates of varying posterior root stimulation frequencies (2.5–50 Hz) in a study of 10 motor complete SCI subjects stimulated while lying supine (Minassian et al., 2004). 2.2 Hz resulted in compound muscle action potentials with short latencies reflecting the activations of group-Ia primary spindle afferents in the posterior root and a subsequent monosynaptic activation of motor neurons. Higher frequencies, however, were suggested to modify the central state of spinal circuits. 5–15 Hz stimulations elicited sustained tonic extensions, while 25–50 Hz stimulations elicited rhythmically alternating flexion/extension activity. Stimulation at both ranges led to lower limb stepping-like movements indicating a cooperated recruitment of different muscles. Transitional frequencies of 5–26 Hz, mapped in another study of 8 subjects with motor complete SCIs, were found to elicit periodic amplitude modulation of lower-limb output EMG patterns (Hofstoetter et al., 2015). Such reflex amplitude modulation consisted of alternations between large and small amplitude responses optimally observed at 16 Hz stimulations and when stimulation was applied to pairs of related muscle groups. The authors proposed different candidate mechanisms centered around synaptic summation of inhibitory and excitatory actions with offset time constants. In addition to this frequency dependent mapping, additional input-output dynamics were identified in 10 subjects with chronic motor complete SCI (Danner et al., 2015). Stimulation induced EMG responses from lower limb muscles were acquired from patients lying supine. Concurrent data from four muscle groups in the same limb showed differential patterns of muscle co-activations, mixed-synergies, and locomotor-like configurations based on input properties.

Given the aforementioned works, it is suggested that (1) EES leads to direct depolarization of large diameter afferents, which subsequently activate both lumbar interneurons involved in lower limb motor control and motor neurons (via mono and poly-synaptic connections) and (2) that stimulation enhances motor activity by increasing the overall central state of excitability lending to an immediate enhancement of motor function when combined with treadmill stepping (Minassian et al., 2007). In line with the proposed synaptic mechanism, computational modeling of alternative lumbar pathways could reproduce some fundamental aspects of the aforementioned frequency dependent responses (Jilge et al., 2004). The recruitment of oligosynaptic pathways at low frequencies *versus* multi-synaptic pathways at higher frequencies was achieved in the model with the addition of an interneuron gate enabling multi-synaptic pathway activation, which opens with temporal summation of successive inputs arriving at short intervals (higher frequency). Altogether, this work pointed to the potential for EES to influence motor function after SCI, as well to the non-linear input-output computations occurring at the level of the spinal cord circuitry with minimal supraspinal influence.

Clinical outcomes from EES combined with training

Work investigating the synergistic effects of stimulation with movement points to a therapeutic value for EES in recovery of motor function in patients with SCI. From an electrophysiological standpoint, examination of EMG rhythmic activity with spinal stimulation alone *versus* spinal stimulation combined with treadmill stepping revealed that central spinal based EMG rhythm differs from that recruited with step-related sensory feedback (Minassian et al., 2013).

Clinical benefits of EES were reported in 2002. A patient with chronic quadriplegia who was wheelchair dependent from an incomplete SCI (Grade C, American Spinal Injury Association) benefited from combined partial weight-bearing therapy and EES (0.8 ms long pulse durations, 20–60 Hz, paresthesia/vibration inducing amplitude) over the upper lumbar enlargement (Herman et al., 2002). Herman et al. (2002) found that longer pulse durations were essential and relatively lower sensitivity to the frequency of choice. While therapy alone was beneficial for improved stereotypic stepping patterns and spasticity reduction, it was insufficient for over-ground stepping (measured by safety, energy cost and fatigue) until coupled with EES. With continuous EES, the patient was able to walk short and long distances (15 m to 50–250 m) with improved gait, doubled speed and increased endurance (8/10 to 2/10 Borg scale) lending to the ability to perform at-home and in-community functional tasks. The authors of the study discussed the possibility that EES augments use-dependent plasticity created by partial weight-bearing therapy and propose this as a dual therapy for individuals with incomplete SCI (Carhart et al., 2004). A subsequent study reported outcomes of a man who was paraplegic for over a 3-year period with complete loss of voluntary motor function yet some preservation of sensory function after a 3-month combined training-stimulation period. The patient achieved full weight-bearing (with balance assistance), locomotor-like EMG patterns, and supraspinal control of some leg movements when training was combined with stimulation *versus* training sessions alone (Harkema et al., 2011). Altogether, these two studies speak to the therapeutic potential of epidural stimulation in individuals with incomplete and complete injuries; authors proposed the revival of previously silent spared spinal circuits and the promotion of use-stimulation dependent plasticity.

Independent stepping enabled by combined stimulation and training in an individual with complete SCI was reported more recently. Initially, with EES, a patient with a complete SCI was able to stand and gain intentional control of step-like leg movements when side-lying or suspended with a body-weight support system (Grahn et al., 2017). However, in a later study, Gill et al. (2018) demonstrated that in the same subject, task-specific training referred to as multi-modal rehabilitation (subject trains to initiate and perform activities in particular positions: laying supine, side-lying, seated, standing, and stepping with trainer assistance and body weight support as needed) combined with epidural stimulation resulted in independent standing, bilateral stepping on a treadmill, stepping over ground (with a front-wheeled walker and trainer assistance for balance), and differential sensorimotor

engagement for stepping *versus* standing. This work demonstrates the possibility of recovering supraspinal-spinal functional networks and consequent motor activity with coupled EES and multiple motor task training.

Additionally, as demonstrated by Angeli et al. (2014), EES can be used to restore conceptual, auditory, and visual input and processing to patients with complete paralysis, restore fine voluntary control upon verbal, auditory and visual inputs. More specifically, lumbosacral EES at 25 or 30 Hz in four individuals for at least 2.2 years post-SCI enabled voluntary lower limb movement upon verbal command. By contrast, no voluntary lower limb movement occurred following verbal command or visual cue in the absence of EES. Furthermore, each of the four individuals were able to achieve normal activation and movement in the ankle and toe muscles via EES. The study concluded that the key factor involved in restoration of lower limb movement to four patients with complete paralysis following SCI was the use of EES to modulate the sub-threshold (near but below motor) state of motor excitability. More specifically, it was found that patients exhibited different stimulation intensity thresholds for movement generation, and that a specific stimulation intensity could evoke different responses: for example, the more spontaneous, spastic patients exhibited movement at lower stimulation thresholds than patients with less spasticity. These results suggest that appropriate EES frequency can help neurons behave according to their normal activation threshold, and that the state of dysregulation in SCI can be attributed to altered spinal cord excitability, which EES may be able to correct. In a follow-up study, it was demonstrated that sub-threshold lumbosacral EES enhanced standing balance in four patients with chronic SCI, one of whom was also included in the prior study (Rejc et al., 2015). The patients with sensory and motor complete SCI progressed to full weight-bearing standing when treated with EES without the need for external assistance. Finally, the study confirmed a previously reported result: projecting weight bearing proprioceptive and/or sensory input to the spinal cord is required for generation of appropriate EMG patterns necessary to enable full-weight bearing standing during EES. Furthermore, in 2017, the same group of researchers investigated the use of EES, again in a group of four patients with SCI. This time, they assessed the effects of EES with standing and subsequent step training on progression of motor function (Rejc et al., 2017a). Ultimately, it was found that stand training yielded inconsistent results in the group of cohort of four patients and that step training following stand training significantly reduced standing ability in three of the patients. From these results, it was concluded that stand and step training with EES does not lead to motor improvements necessary for standing, and that step training may induce nervous system alterations that interfere with standing ability.

Motor training with EES was further investigated by Rejc et al. (2017b) who studied the progression of voluntary leg movement and standing (without EES) in a patient with C7 level injury who underwent 3.7 years of an EES, motor task-based intervention. The tasks trained during EES were standing, stepping, and volitional leg movement, and performance of these motor tasks without EES improved over the course of the study. In fact, the patient was unable to perform knee extension or hip flexion without EES prior to the intervention, but could perform volitional leg movements with EES following only four days of intervention. This case was significant in that it was the first report demonstrating neural adaptation-mediated improvement from zero volitional motor ability to significant volitional lower limb control and unassisted standing ability (both in absence of EES) following treatment with 2 Hz EES. The potential motor mechanisms involved in the adaptation may have included strengthening of residual descending pathways through plasticity and or plasticity-mediated improvements in local spinal circuits. In certain cases, these improvements are likely garnered from neuromodulation of residual, or subfunctional neural circuits presents in the spinal cord, as demonstrated by a 2017 study out of Mayo Clinic which built on the results obtained by Rejc et al. (2017b). The authors showed that lumbosacral EES led to improved volitional control of task-specific and rhythmic (steplike movement) motor activities as well as standing (Grahn et al., 2017). This was, in fact, the first report of EES leading to restoration of the ability to perform these tasks in a single patient within 2 weeks of intervention onset (of note, without EES, the patient was unable to perform the aforementioned tasks). The results of this study imply that subfunctional neural circuitries may be present in certain clinically motor complete (ASIA A or B) cases of SCI. Hypothetically, these subfunctional neural networks can be targeted for neuromodulation through activity-dependent reorganization and appropriate integration of input from proprioceptive inputs, which are critically involved in post-SCI reorganization (Taccola et al., 2018). As such, future studies should investigate how these pathways can be targeted during EES therapy to restore volitional motor function in SCI.

In order to best restore such function, it is important to understand how EES electrodes can be optimally implanted and positioned in patients. For example, intraoperative electrophysiological techniques can be used to position the electrode: once the electrode array is inserted, signals from recording electrodes placed in muscles of the lower limb can be used to assess EES preferential activation of proximal *versus* distal muscles as well as to monitor symmetry of lower limb activation (Calvert et al., 2019b). Furthermore, subtle alterations in limb position can be guided by EMG recordings of motor responses induced by EES. In this manner, EES-induced motor responses may guide proper placement of EES electrodes for selective targeting of specific circuits in the spinal cord. Specific targeting is important because it can enable EES to selectively stimulate specific posterior roots. In this manner, EES can recruit proprioceptive inputs within the posterior spinal cord roots, which have been found to be central to the ability of EES to engage motor neurons at a particular spinal cord segment (Wagner et al., 2018). More specifically, spatiotemporal EES applied to the posterior roots allows for stimulation to be timed in order to coincide with the desired movement. By contrast, continuous EES can produce movement only when constrained to a limited range of simulation parameters, and does not yield rehabilitation-independent improvements in humans (Formento et al., 2018). This further supports the proposed mechanism of EES stimulation of posterior root neurons and points to the importance of proprioceptive input, which is likely necessary for EES induction of motor-pattern formation. Because EES evokes both pro- and anti-dromic signals in bipolar sensory neurons, continuous EES – which results in a higher occurrence of collisions between normal and EES-induced antidromic signals – may disrupt proprioceptive input. Non-continuous (burst) EES and spatiotemporal EES therefore provide the advantage of reduced likelihood of such cancellation, meaning they better preserve sensory input. As these methods demonstrate superior outcomes over continuous EES, it is likely that EES cannot interfere with normal proprioception if restoration of motor function is to be obtained.

Accordingly, great improvements in EES capabilities have come with fine-tuning the execution of this technology. In 2018, a study reported restoration of intentional over-ground walking ability in two of four patients with chronic motor complete spinal cord injury (Angeli et al., 2018). The discordant outcomes among the four patients may have been the results of sensory sparing, the role of which is currently under study. Furthermore, it is notable that successful walking was achieved only in the presence of EES and the patient's

intent to walk, which suggests that EES activates interneuronal lumbosacral spinal networks through dorsal nerve roots or stimulation of the spinal cord parenchyma. Additionally, rhythmic activation of lower limb muscles appeared to be in tune with the step cycle and did not directly correlate with stimulation frequency, which suggests that EES was responsible for activation of multiple groups of neurons within the spinal cord.

As EES continues to be optimized and its mechanisms of neuromodulation in restoring lower limb function uncovered, it has also demonstrated potential for restoration of upper limb activity in tetraplegic SCI patients. However, it is important to note that the nature of neuromodulation of the upper limbs may differ from that of the lower limbs due to differences in neuromuscular properties. For example, volitional control of upper limbs is less "automatic" than maintenance of posture and locomotion (Lu et al., 2016). Regardless, EES of the cervical spine has demonstrated the ability to restore hand motor function in tetraplegic patients. For example, EES was used to improve volitional hand control and grip strength in two patients with chronic cervical SCI. Even following a single EES session, grip strength, motor control, and action latency were improved, and long-term improvements in hand (three-fold increase in grip strength) and arm motor function were observed in both patients over the course of treatment. Furthermore, the patients exhibited similar improvements despite the fact that they had experienced different injuries and had undergone different procedures for these injuries. As restoration of upper limb function could significantly improve the quality of life of tetraplegic patients, future studies should be conducted to determine how EES can be used to modulate the neuronal excitability and functionality of spinal cord neurons to best facilitate motor function.

Limitations of Epidural Electrical Stimulation

A factor potentially holding back EES studies is that it is challenging to eliminate bias that is seemingly intrinsic to the study design (Darrow et al., 2019). For example, in cases where study "assessors" and participants are aware of current changes, blinding is impossible to achieve (Darrow et al., 2019). Unfortunately, most studies investigating EES up to this point have included case series and case studies, such that outcomes are limited to those obtained from cohorts of only several patients at a time. At present, although EES has demonstrated the ability to restore motor function in patients with SCI, it has yet to achieve the regulatory requirements necessary for FDA approval (Calvert et al., 2019a). As such, the current challenge facing researchers is the necessity to integrate EES into clinical efforts that can potentially establish its efficacy for SCI. Two potential avenues exist by which EES could gain FDA approval. The standard clinical trial phase route could be pursued, or an exemption for a particular treatment could be obtained in order to garner FDA approval (Youngerman et al., 2016).

Besides the logistical limitations associated with studies investigating EES, there are significant questions related to the procedure and technology itself that must be addressed before translation to the clinic can be achieved. For example, spinal procedures such as laminectomy are currently required for placement of the EES device (Calvert et al., 2019a). These procedures come with bio-mechanical risks such as spinal cord instability and/or deformity as well as the risks associated with surgery in general (infections, blood loss, etc.). Clearly, a less invasive placement method is desirable, but the reality is that the vertebral lamina must be removed in order to gain access to the spinal cord. Besides the procedural drawbacks of EES, another limitation is the heterogeneity of modulation parameters that have been deemed effective for specific motor functions in individual patients (Calvert et al., 2019a). As a result of this limitation, great time is required to achieve effective EES for different patients, each of whom present with unique injuries and spinal cord anatomies. The diverse array of configurations and stimulations parameters required for motor function across patients is an immense collection of different frequencies, placement locations, and other settings. This heterogeneity makes results – some of which may not be applicable to the next patient - difficult to interpret; this is particularly unfortunate consider the great challenge required to arrive at optimal stimulation parameters for a particular patient. Another factor potentially confounding result is the fact that both physician rehabilitation and EES likely play a role in functional motor recovery. Separating out of the role of EES alone remains difficult, especially because studies have been designed to demonstrate lack of recovery in motor rehabilitation alone (before the application of EES with motor rehabilitation) (Calvert et al., 2019a). For this reason, and because EES motor rehabilitation must continue to be optimized, future studies should investigate which forms, frequencies, and volumes of motor training yield the best results when coupled with EES.

Concluding Remarks

As we have discussed in this review, significant progress has been made to elucidate the pathophysiology of SCI and employ EES in the recovery of motor function. By modulating the spinal neural network, EES has been able to restore stepping abilities in animal models and patients with SCI. Notably, recent studies with human subjects have demonstrated that targeting proprioceptor circuits plays a crucial role in the reorganization of residual descending pathways. Future directions for EES for SCI will need to overcome several challenges. For example, refinements will be required to establish the optimal EES techniques and stimulation parameters. Improvements in the biocompatibility of electrodes will be essential to minimize long-term complications. Further challenges include conducting studies with a large subject number to evaluate the efficacy and safety of EES. Although challenges remain, groundbreaking cases demonstrating the efficacy of EES for SCI are emerging. Thus, interdisciplinary collaborations will enable the mechanistic understanding of EES in the restoration of the neural circuit and accelerate the implementation of this strategy in clinical settings.

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